October 7, 2014

Continue testing sensor

- Transform plasmids into Top10 Cells
- Overnight cultures - dilute 100 µl in 2 ml
    - Grow at 37 °C for 1.5 hours
    - Add 1 µl of beta-estradiol in ethanol (10 mg/ml)
        - cultures treated with ~20 µM of estrogen
- Incubate at 30 °C
- Grow overnight

October 8, 2014

Continue testing sensor

- Set up cultures of WT sensor, DEAD Sensor, 115 YFP, 115 YFP and T7RFP term, T7RNAP YFP and T7RFP term, and Top10 cells only at 37 °C, 30 °C and 20 °C to grow overnight

Transformation of Killer Red Codon Optimized and Supernova Codon Optimized into Top10 Cells

- 100 µl of cells for remaining DNA (about 1 - 3 µl)
- Plate 400 µl transformants on LB + Cam plates

iGEM Transformation Protocol: http://parts.igem.org/Help:Protocols/Transformation

October 9, 2014

Continue testing sensor

- Cells at 20 °C grew too slowly to be read
- Cells at 30 °C and 37 °C read in TECAN at yellow and red wavelengths

Table 1. TECAN Parameters used to Read Samples

Fluorescent Protein	**Excitation Wavelength (nm)**	**Emission Wavelength (nm)**	**Gain (if not optimal)**
YFP	514	527	90
RFP	584	607	175

Set up cultures of 115 YFP, 115 YFP and T7RFP term, T7RNAP YFP and T7RFP term in triplicate at 37 °C, 30 °C and 20 °C to grow overnight